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Positive Responses of Nitrogen-Fixing Bacteria Combined with Vermicompost on Farmland Heath and Peanut Yield

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A study on the supplementation of *Bacillus vir*eti combined with the vermicompost with two types of irrigation water [river water (RV) and underground water (UW)] on soil properties, growth, yield, and arsenic (As) accumulation of peanuts. The field study that was performed in Phuoc Hung, An Giang, Vietnam, consisted of three factors: (1) two vermicompost rates (0.0 and 10 t/ha); two inoculated methods of *Bacillus vireti* (supplementation and no supplementation); (3) two irigation water types (RW and UW) and four replications. The novel findings of this study include (i) a significant increase in fresh peanut yield, ranging from 11.0% to 23.4%, when 10 t vermicompost /ha was applied in combination with *Bacillus vireti* and non-As contaminated irrigation water (RW) compared to the control; (ii) a significant reduction in As accumulation in both stems (9.96% to 39.2%) and seeds (12.9% to 30.2%) due to the positive effects of *Bacillus vireti*. These results suggest that *Bacillus vireti* species has the potential to enhance soil fertility, peanut yield, and reduce As accumulation in peanuts when combined with vermicompost application. Moreover, this is a promising nitrogen-fixing microorganism for the production of organic microbial fertilizers to enhance soil health and crop productivity in the future.
Keywords: Animal manure, deep well water, endophytic bacteria, groundnut, soil nutririon, supplementation.

INTRODUCTION

Groundnut is an oilseed and an important industrial crop worldwide. In addition, peanuts are a soil-improving crop due to their ability to fix nitrogen, and they are also an important food crop and oil crop among annual oilseed crops. Peanut by-products are used as animal feed and fertilizer. The local farmers have a long tradition of peanut production in An Giang (Nguyen et al., 2024; Sobolev et al., 2013). Various bacteria that interact with the peanut roots, often associated with N-fixing and alleviating plant (Chen et al., 2019). Several fungal and bacterial diseases, which include leaf spot, stem and pod rot, root rot, and wilt, commonly occur in peanut cultivation. Nitrogen-fixation microorganisms in plant roots are considered a great source of biofertilizers and excellent biocontrol agents in agricultural cultivation (Sobolev et al., 2018). Nitrogen-fixation microorganisms that increase in the soil and plant health, also transfer these positive abilities from this generation to the others (Ludueña et al., 2018). Nitrogen-fixation microorganisms which included nitrogen-fixing bacteria have the ability of growth promotion and symbiotic nitrogen fixation relationship with peanut plants (Prestes et al., 2019). The rhizosphere endophytic bacteria influence plant growth and defense by producing a variety of phytohormones (Fincheira & Quiroz, 2018). Vermicompost application and *Bacillus vir*eti inoculation on low nutrient soil significantly increased yield components, yield, and peanut quality under outside net house conditions at the An Giang University (AGU) Experimental Center. The study revealed the effect of *Bacillus vir*eti species on nodule number and peanut yield in the prior crop. In the next crop, the previously experimental plots of vermicompost application and endophytic inoculation significantly increased plant height, biomass, and number of pods, nodules, yield, and peanut seed quality compared to no vermicompost application and nitrogen-fixing bacteria in prior crop (Chuong & Tri, 2024). The As accumulation in farmland soils has gradually hoarded from the polluted resources of irrigation water and chemical fertilizers, which have significantly concern the pollued environment, agricultural products, and human health

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because of As toxicity. Even low-level exposure to As can alter plant morphology, physiology, and biochemistry (Van, 2024; Abbas et al., 2018). Yields of peanut and other crops have significantly declined in recent years in Vietnam, primarily due to infertile soil and high As content in soil and water (Nguyen & Tran, 2024). The main cause reduced peanut profitability for local farmers, who was the excessive use of chemical fertilizers and high fertilizer prices, coupled with low peanut yields. Therefore, improving this issue involves applying organic fertilizers in combination with NFM strains to enhance peanut quality and yield (Nguyen, 2024). NFM have the ability to manufacture the natural nitrogen fertilizer from the air around the peanut root areas (Chuong, 2024a). The vermicompost application has been shown to increase peanut yield and improve soil fertility (Getachew et al., 2019). Using organic manures to immobilize As toxins in contaminated soil has been a well-established and widely used technology. The application of inorganic and organic amendments, which can immobilize heavy metals and As toxicology in soil, leads to increased crop yield and soil fertility (Chuong, 2024b; Chaw et al., 2018). Increasing peanut yield and reducing As content in soil can contribute to improve soil nutrients and NFM density through fertilization and vermicompost addition (Zhu et al., 2018; Chuong, 2024a). In recent years, research on endophytic microorganisms has demonstrated their ability to enhance nutrient uptake, stress tolerance, and disease resistance in host plants, leading to improved crop yields. Evidence suggests that endophytes can enhance tolerance to salinity, heavy metal contamination, and drought conditions (Watts et al., 2023). Furthermore, endophytes provide a sustainable alternative to traditional agricultural practices, reducing the need for synthetic fertilizers and pesticides, thereby mitigating the risks associated with chemical treatments. Studies have also shown that combining endophytes with organic fertilizers in agriculture is a key factor in achieving sustainable agricultural productivity and maximizing their potential (Liu et al., 2023; Chuong, 2024b). Given its exceptional adaptability to sandy soils, peanut cultivation has emerged as a highly profitable agricultural cultivation for regions characterized by limited irrigation, particularly in hilly terrains devoid of adequate irrigation infrastructure. Agricultural scientists have been instrumental in guiding farmers towards a sustainable cultivation model that integrates nitrogen-fixing microorganisms and organic manures, thereby significantly improving soil fertility and consequently, peanut yields (Chuong, 2024c). The study goal was to appraise the impact of vermicompost and *Bacillus vireti* supplementation on soil fertility and peanut yield grown on As-contaminated soil and irrigation water. The study findings could provide local farmers with relevant information advising them to utilize vermicompost and *Bacillus vireti* supplementation in conjunction with uncontaminated As irrigation water to boost peanut yield and reduce As content in agricultural products.

MATERIALS AND METHODS

***Bacillus vireti isolation and identification*:** *Bacillus vireti* species that isolated from peanut nodules at 65 days after planting (Hossain et al., 2023). All fifty selected nodules from five plants (ten nodules/plant) that were well washed with fresh water in order to clean the nodules, were carefully rewashed on nodule's surface by 70% alcohol, and then, rewashed with pasteurized distilled many times, steeped in 5% NaCl for 1 minute, finally. The cleaned nodules were taken in 1 mL of phosphate-buffer, and streaked on YMA nutrition plates. The YMA plates were diluted with many different rates and the streaked YMA plates, which were incubated at room temperature about four days. The pure colonies were selected for molecular identification. *Bacillus vireti* was extracted to collect DNA from pure colonies for 16S rRNA gene analysis to compare with the similarity of previously designed bacterial sequences. *Bacillus vireti* was isolated in the laboratory of An Giang University (Figure 1). The 16S rRNA gene was amplified from these isolates using the 27F and 1492R primer pair in three PCR cycles. The DNA sequence similarity of these isolates further determined belong to previously published sequences of nodulose bacteria (Cardoso et al., 2018). NCBI BLAST technology was used to interpret the Sanger sequencing results, and the selected isolate branch with high similarity up to 100% to the *Bacillus vireti* species based on sequencing results and phylogenetic tree (Figure 2). The *Bacillus vireti* density was sufficiently increased to 10^8CFU/mL five days before sowing, and then, co-inoculated with peanut seeds and soaked in liquid YMA medium for 8-12 hours before sowing (Chuong & Tri, 2024; Etesami, 2022).

***Experimental design and location*:** An Phu District is located on an island with a total area of 225.3 km^2 and has up to 200,000 people. It borders Cambodia. The annual rainfall ranges from 500 to 650 mm, with 60 to 70% of it falling between June and October. The average temperature ranges from 27 to 38°C (summer) and from 20 to 32°C (winter).

The field experiment that consisted of eight treatments inside the dike and arranged in a completely randomized block design in Phuoc Hung commune, An Phu district, An Giang province from Eight to November of 2024, had 4 replications. The eight treatments, which were coded from 1 to 8, consisted of: (1: control) UW irrigation (without *Bacillus vireti* and vermicompost); (2) RW irrigation (without Bacillus vireti and vermicompost); (3) appliction of 10t vermicompost/ha and UW irrigation (without *Bacillus vireti*); (4) application of 10t vermicompost ha^{-1} and RW irrigation (without *Bacillus vireti*); (5) *Bacillus vireti* supplementation and UW irrigation (without vermicompost); (6) *Bacillus vireti* inoculation and RW irrigation (without vermicompost); (7) *Bacillus vireti* supplementation, 10t vermicompost ha^{-1} amendment, and UW irrigation, (8) *Bacillus vireti* inoculation, 10t vermicompost ha^{-1} amendment, and RW irrigation. Three

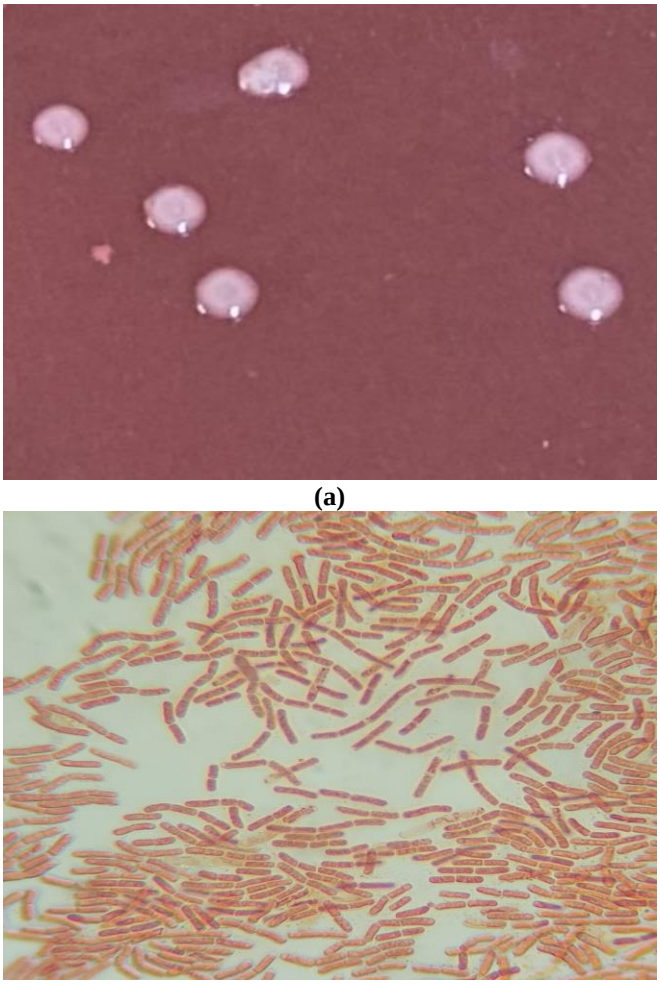
factors consisted of (factor 1) *Bacillus vireti* (supplementation and no supplementation)*;* (factor 2) irrigation water types (RW and UW); (factor 3) 10 t vermicompost/ha amendment and no vermicompost amendment.

The chemical fertilizers were utilized by single fertilizers, which included urea, superphosphate, and potassium chloride. Then, the amounts of these fertilizers were converted to kilograms of N, P, and K per hectare, which were applied by 40N:60P: 60K kg/ha for eight experimental treatments. Vermicompost that was collected by Nongnghieppho company, Vietnam, contained total compositions of N (0.98%), P (1.05%), kali (0.29), Ca (1.18%) and Mg (0.27%). The whole area of the experiment was 640 m^2 (1 m in width x 20 m in length x 8 plots x 4 replication). The spacing between two plots was 0.5 m and were planted in single rows with a spacing of 25 cm between plants (sowing 2 seeds/hole). When the plants had a maximum of 3 leaves, one healthy plant was selected from each hole for monitoring throughout the study. The initial chemical properties of the RW were pH (5.7), while the UW was pH (4.7). The As concentration that was not detected in RW, contrary, was present in UW at a concentration of 697 µg/L. The WHO guideline for arsenic in irrigation water is 50 µg/L. The WHO guideline for As content in irrigation water is 50 µg/L. As such, the As concentration in the UW in the study area was 10 times higher than the WHO guideline. Prolonged drought caused by climate change has led to the RW shortage for crop irrigation in An Phu district, An Giang, Vienam. Local farmers have been forced to use UW, which contains high As content to irrigate their crops, results in a remarkably decrease in crop yields and quality (Chuong, 2024b). The initial soil properties were pH (4.97), CEC (2.627 $cmol^+/kg$), soil organic matter (SOM:1.24%), total nitrogen (N:0.125%), available phosphorus (P:25.4 mg/kg), and exchangeable potassium (K:75.1 mg/kg). The arsenic concentration in the soil was 84.55 mg/kg (inside the dike). The experimental soil texture was silt sandy loam with silt (35.1), sand (58.5%) and lay (6.4%), which were evaluated according to (USDA, 2019) and suitable for peanut growth (Van Chuong & Le Kim Tri, 2024; Chuong et al., 2024).

***Data collection*:** (i) The plant height, chlorophyll concentration, branch and leaf number (No.) were averagely measured per plant during 20, 45 and 65 DAP. Each treatment selected ten plant to observe growth and yield traits, which included the nodulose No. and Weight (Wt.) of fresh nodulose and biomass, No. of full and empty pods, Wt. of 1,000-seeds and fresh pod yield; (ii) nutrition composition of seeds: seed samples were further determined for humidity, lipid, protein; (iii) As toxicology: Samples of trunks, leaves and seeds were taken per replication at harvest season and dried separately for As determination.

***Analysis methods*:** Samples of soil, trunk, leaf, and seed were separately taken and dried to Analyze As, exchangeable K and grain nutrient compositions. Arsenic concentrations of soil, stems, and seeds were analyzed by Korkmaz et al. (2017). Soil samples were collected from each replication to determine some soil chemical properties. pH meter was measured soil pH with 1:2.5 ratio of soil-to- water. Total nitrogen concentration was determined by the Kjeldahl method, while available P was used by the alkaline hydrolysis method, as outlined by All parameters were analyzed by using the USDA method.

***Statistical analysis*:** Statgraphite XV software was used for the variance analysis (ANOVA). The mean comparison between variables was performed by Duncan's test with a significant difference at *P value* ≤0.05. Each treatment's data were calculated according to 3 factors to find the interaction between the experiments and the factors.



(a)

(b)

Figure 1. (a) colony isolation of *Bacillus vireti* on YMA medium; (b) Scanning 100X microscope showing cell morphology of *Bacillus vireti.*

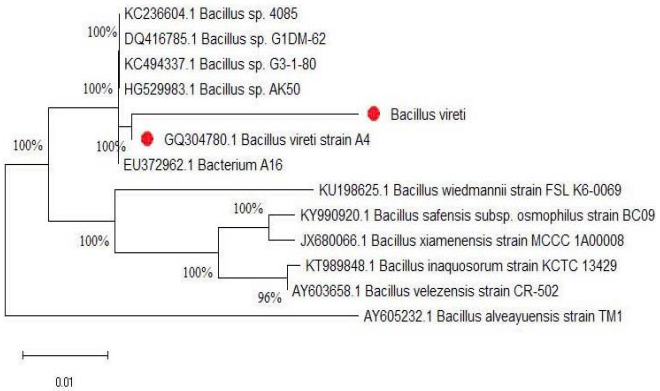


Figure 2. Phylogenetic tree of *Bacillus vireti* species along with the 16S rRNA gene sequences from choosen references strains, which was constructed by using MEGA 11. The scale bar corresponded to 0.01 per nucleotide site. The node number are based on 1,000 replications of bootstrap values.

RESULTS

***Soil chemical properties at harvest*:** Table 1 proclaims that application of 10 t vermicompost/ha was highly significant differences (p ≤0.01) on soil pH, CEC, SOM, total N and available P. The chemical traits of the farmland in the treatments of 10 t vermicompost/ha fertilization were all higher than the control treatments. Similarly, the *Bacillus vireti* supplementation increased in all soil nutrient properties at harvest season compared to the non *Bacillus vireti* supplementation (except for total N) and was highly significant differences (P≤0.01). Two irrigation water types, the use of As-contaminated UW for watering peanut plants resulted in lower soil nutrients than RW irrigation, and were statistically significant differences (P≤0.01). Meanwhile, plots of vermicompost application, *Bacillus vireti* supplementation and two irrigation water types had very large interactions between them (P≤0.01). Total N did not interact between the experiments without the *Bacillus vireti* supplementation. Furthermore, the total N amount was insignificantly different between the vermicompost application and *Bacillus vireti* supplementation (Table 1). Soil pH valued from 5.14 to 6.10 at the factors of vermicompost, *Bacillus vireti* supplementation and irrigation water types at harvest season. All soil pH values in 10 tons of vermicompost, *Bacillus vireti* supplementation inoculation, and RW irrigation were all higher than others. Similarly, values of the CEC, SOM, total nitrogen, and available phosphorus in the experiments with 10 t vermicompost/ha, *Bacillus vireti* supplementation, and RW irrigation were all higher than the treatments without vermicompost application, without *Bacillus vireti* supplementation, and with As-contaminated UW irrigation. However, the value of total nitrogen was not statistically different between *Bacillus vireti* supplementation and no *Bacillus vireti* supplementation. In this study, the vermicompost application (factor A), *Bacillus vireti* supplementation (factor B), and two irrigation water types (factor C) had a highly significant effect (p ≤ 0.01) on the chemical components of the soil, and the interaction between the factors also had a significant effect (P ≤ 0.01).

***Peanut Agronomy components*:** Plant height is an important trait for evaluating peanut plant development, was observed during period of 20, 45, and 65 DAP (Table 2). Efficiency use of soil nutrition and disadvantageously environmental sources. As shown in the Table 2 proclaims that the individual influence of vermicompost application, Bacillus vireti supplementation, and two irrigation water types on plant height at three growth periods of peanuts were significantly differed by 1% level, on contrary, their interactions did not differ significant for this component [Except for the interaction between vermicompost application (A) and

Table 1. Chemical properties of soil at harvest season.

Factors	pH	CEC cmol$^+$/kg)	SOM (%)	Total N (%)	P(mg/100g)
		Vermicompost (A)			
0.0 t/ha	5.14**±0.03^b	6.09**±0.04^b	1.18**±0.02^b	0.09**±0.01^b	20.8**±0.08^b
10 t/ha	5.90**±0.08^a	6.61**±0.01^a	2.10**±0.08^a	0.14**±0.03^a	32.9**±0.08^a
		***Bacillus vireti* supplementation (B)**			
Yes	6.00**±0.41^a	7.56**±0.04^a	1.59**±0.01^a	**0.11±0.01^{ns}**	29.6**±0.16^a
No	5.24**±0.03^b	5.05**±0.04^b	1.50**±0.08^b	0.10±0.40^{ns}	25.9**±0.08^b
		Irrigation water types (C)			
UW	5.34**±0.03^b	5.23**±0.03^b	1.99**±0.08^b	0.08**±0.01^b	30.5**±0.42^b
RW	6.10**±0.08^a	7.98**±0.01^a	2.21**±0.08^a	0.18**±0.07^a	35.2**±0.16^a
F (AxB)	**	**	**	ns	**
F (AxC)	**	**	**	**	**
F (BxC)	**	**	**	**	**
F(AxBxC)	**	**	**	**	**

ns: no significant difference at P value > 0.05, **: significant difference at P value ≤ 0.01

Table 2. Effect of *Bacillus vireti,* vermicompost and irrigation water on agronomic traits.

Factors	Average height per plant (cm)			Available branch number (branches/plant)		
	Days after planting (DAP)					
	20	45	65	20	45	65
Vermicompost (A)						
0.0 t/ha	11.1**±0.01^{b}	20.4**±0.33^{b}	49.3**±0.25^{b}	3.75**±0.04^{b}	4.50**±0.41^{b}	5.12**±0.02^{b}
10 t/ha	12.7**±0.16^{a}	22.8**±0.16^{a}	53.2**±0.16^{a}	3.86**±0.16^{a}	5.75**±0.04^{a}	6.25**±0.04^{a}
***Bacillus vireti* supplementation (B)**						
Yes	13.0**±0.81^{a}	23.1**±0.08^{a}	53.7**±0.16^{a}	3.67**±0.02^{a}	5.50**±0.41^{a}	6.50**±0.41^{a}
No	10.6**±0.16^{b}	19.6**±0.16^{b}	48.4**±0.33^{b}	3.50**±0.41^{b}	4.75**±0.04^{b}	5.50**±0.41^{b}
Irrigation water types (C)						
UW	9.90**±0.08^{b}	18.6**±0.16^{b}	46.7**±0.16^{b}	3.50**±0.41^{b}	5.12±0.02^{ns}	5.69*^{s}±0.46^{b}
RW	12.4**±0.33^{a}	22.1**±0.08^{a}	51.9**±0.08^{a}	3.80**±0.08^{a}	5.13±0.02^{ns}	5.88*^{s}±0.07^{a}
F (AxB)	ns	ns	ns	**	**	**
F (AxC)	ns	**	**	ns	ns	ns
F (BxC)	ns	ns	ns	**	**	**
F (AxBxC)	ns	ns	ns	**	**	**

ns: no significant difference at P value > 0.05, *; **: significant difference at P value ≤ 0.05 and ≤ 0.01, respectively

Table 3. Effect of *Bacillus vireti* and irrigation water on the yield traits at harvest.

Factors	Biomass (gr/plant)	No. of full pods (pod/plant)	full pod Wt. (gr/plant)	Nodule No. (Per plant)	nodule Wt. (gr/plant)	Wt. of 1,000 seeds (gr)
Vermicompost (A)						
0.0 t/ha	244**±2.00^{b}	63.8**±0.65^{b}	158**±1.60^{b}	235**±4.08^{a}	0.96**±0.02^{b}	860**±8.16^{b}
10 t/ha	282**±1.63^{a}	78.3**±0.25^{a}	176**±0.96^{a}	175**±4.08^{b}	1.27**±0.02^{a}	1,050**±4.08^{a}
***Bacillus vireti* supplementation (B)**						
Yes	296**±1.63^{a}	80.8**±0.65^{a}	175**±0.82^{a}	257**±1.63^{a}	1.39**±0.01^{a}	1,050**±4.08^{a}
No	228**±1.63^{b}	64.9**±0.08^{b}	135**±0.82^{b}	155**±4.08^{b}	1.22**±0.02^{b}	840**±8.16^{b}
Irrigation water types (C)						
UW	244±3.26^{ns}	78.3ns±0.245	131**±0.82^{b}	181**±0.82^{b}	0.90**±0.08^{b}	870**±4.08^{b}
RW	260±4.08 ^{ns}	76.5ns±0.408	171**±0.82^{a}	307**±1.63^{a}	1.70**±0.08^{a}	925**±2.88^{a}
F (AxB)	**	**	**	**	**	**
F (AxC)	**	**	**	ns	ns	**
F (BxC)	ns	**	**	**	**	**
F (AxBxC)	**	**	**	**	**	**

ns: no significant difference at P value > 0.05, **: significant difference at P value ≤ 0.01

irrigation water types (C) at the 45 and 65 DAP at the 1% level]. The plant height of 20, 45 and 65 DAP in all experiments, which were applied with 10 t vermicompost/ha, Bacillus vireti supplementation, and RW irrigation, were higher than others. The number of available branches at growth periods of 20, 45 and 65 DAP (P≤0.01) was statistically different between treatments at factor A (vermicompost) and factor B (Bacillus vireti). However, stages of the 45 and 65 DAP were insignificantly differed by the 1% level for two irrigation water types (expect for 20 DAP). Irrigating with RW (non-As-contaminated water), vermicompost application and Bacillus vireti supplementation had more available branches than the control plots of non-vermicompost application, non Bacillus vireti supplementation, and UW irrigation. There was significant interaction between three factors (vermicompost application, Bacillus vireti supplementation and two irrigation water types) at period of 20, 45, and 65 DAP at the 1% level [except (AxC)]. Application of organic manures, combined with NFM inoculum and As-free irrigation water helped peanut plants to produce more effective branches, leaves, and flowers.

***Peanut yield traits*:** The peanut biomass was affected by application of vermicompost and Bacillus vireti supplementation at the 1% level [expect Factor (C)] (Table 3). All treatments of vermicompost application combined with Bacillus vireti supplementation had higher peanut biomass than the control treatments [expect Factor (C)]. The interaction of the experimental factors on peanut biomass was significantly different at the 1% level [expect Factor (BxC)]. The Bacillus vireti supplementation had the highest biomass (296 gr/plant) and the treatment only applied NPK and As-

contaminated irrigation had the lowest biomass value (244 gr/plant) (Table 3). The number and weight of full pods and nodules that consisted of factor A (vermicompost); factor B (Bacillus vireti supplementation) and factor C (irrigation water types), were significantly differed among treatments at the 1% level (P value ≤ 0.01). There were also significantly statistical differences at 1% among their interactions, except F(AxC) at number and weight of peanut nodules. There was significantly statistical difference at the 1% level for weight of 1,000 seeds at F(AxBxC). Treatments of vermicompost application associated with Bacillus vireti supplementation and RW Irrigation resulted in higher weight of 1,000 seeds than the control treatment of without vermicompost application, no Bacillus vireti supplementation and UW irrigation.

***Nutrition composition and As content of peanut seeds*:** Different vermicompost rates were significant affected on humidity percentage of peanut seeds (Table 4). The plots of vermicompost application (32%) obtained the higher moisture of without vermicompost application (30%), and the *Bacillus vireti* supplementation (32.8 %) provided the higher humidity percent of non *Bacillus vireti* supplementation (29.2%), similarly, seed moisture percentage of treatments with irrigating UW (30%) was lower seed humidity percentage of RW irrigation (32.1%). There were significantly differed among treatments at the 1% level in three factors, and their interaction were insignificantly differences. The application of 10 t vermicompost/ha obtained the higher lipid content (25.9%) than compared the lipid content of without vermicompost application was 25.3% (Control) and significant differences at level 1%. The lipid concentration of treatments with *Bacillus vireti* supplementation was insignificantly differed between treatments of factor B (*Bacillus vireti* supplementation). However, the lipid content that obtained at treatment of RW irrigation, was higher than lipid content of UW irrigation. Their interactions of A×B, AxC, BxC and AxBxC were insignificantly differed on seed lipid contents of peanut (Table 4). The interaction among three factors were insignificant differences [except F(BxC) was different at the 1% level]. The different effect was inadequately on the protein percent of peanut seeds in factor A and B (except factor C). The peanut seed protein of UW irrigation was the lower content (15.2%), compared to RW irrigation (19.1%) and the significant differences at level 1%. (Table 4). Nitrogen - fixing Ability of Peanut can meet most of their nitrogen requirements through a symbiotic relationship with NFM strains. The available nutrients are mainly determined by the diversity of microbial resources and their activities in the rhizosphere, which affected on plant health, nutrition, and plant yield, as well as soil quality and soil fertility. The fresh pod yield of three factors valued from 6.03 to 7.87 t/ha, the highest fresh pod yield of peanut was 7.87 t/ha at treatments of RW irrigation, and the lowest fresh pod yield of peanut (6.03 t/ha) at treatments of UW irrigation. The fresh pod yields of 10 t vermicompost/ha application, *Bacillus vireti* supplementation and RW irrigation were higher than treatments of non- vermicompost application, non *Bacillus vireti* supplementation and UW irrigation and statistically significant differences at level 1%.

Table 4 shows that application of 10 t vermicompost per ha had a highly significant effect (p≤0.01) on As accumulation of peanut stems and seeds. The average As concentration of peanut stems and seeds of 10 t vermicompost/ha application was lower than the control treatments (without vermicompost application). Similarly, the factor B (*Bacillus vireti* supplementation) decreased in peanut stems and seeds at harvest compared to non- *Bacillus vireti* supplementation, and was significant differencé (P≤0.01). When considering the irrigation water types, the use of UW irrigation (As-

Table 4. The fresh yield and quality of peanut at harvest.

Factors	Humidity (%)	Lipid (%)	Protein (%)	Fresh yield (t/ha)	As contents (µg/kg)	
					Stems	Seeds
Vermicompost (A)						
0.0 t/ha	30.0**±0.8^{b}	25.3**±0.25^{b}	17.3±0.245^{ns}	6.53**±0.03^{b}	1,064**±3.27^{a}	116**±0.82^{a}
10 t/ha	32.0**±0.8^{a}	25.9**±0.08^{a}	17.4±0.25 ^{ns}	7.34**±0.03^{a}	958**±1.63^{b}	101**±0.82^{b}
***Bacillus vireti* supplementation (B)**						
Yes	32.8**±0.16^{a}	25.8±0.16 ^{ns}	17.3±0.24^{ns}	7.47**±0.02^{a}	944**±3.27^{b}	99.5**±0.41^{b}
No	29.2**±0.16^{b}	25.4±0.33 ^{ns}	17.4±0.33^{ns}	6.40**±0.08^{b}	1,077**±1.63^{a}	118**±1.63^{a}
Irrigation water types (C)						
UW	30.0**±0.82^{b}	23.5**±0.41^{b}	15.2**±0.16^{b}	6.03**±0.02^{b}	1,260**±4.98^{a}	128**±1.63^{a}
RW	32.1**±0.08^{a}	27.7**±0.16^{a}	19.1**±0.08^{a}	7.87**±0.06^{a}	766**±4.89^{b}	89.3**±0.24^{b}
F (AxB)	ns	ns	ns	ns	ns	**
F (AxC)	ns	ns	ns	ns	ns	**
F (BxC)	ns	ns	**	ns	ns	**
F (AxBxC)	ns	ns	ns	ns	ns	**

ns: no significant difference at P $_{value}$ > 0.05, **: significant difference at P $_{value}$ ≤ 0.01

contaminated water) had in higher As accumulation in the stems and seeds than RW irrigation, and were statistically significantly different (P≤0.01). Meanwhile, plots of vermicompost application, *Bacillus vireti* supplementation and irrigation water types did not interact between them in As accumulation of peanut stems. However, their interactions of A×B, AxC, BxC and AxBxC were insignificantly different on As contents of peanut seeds (Table 4).

DISCUSSION

The analysis of soil nutrient traits at harvest season revealed that inoculation with *Bacillus vireti* in combination with vermicompost application, and RW irrigation, resulted in a marked enhancement of soil nutrient content relative to the control (Table 1). The prior research indicated that soil pH, chemical trait, and nutrition of farmland increased after the organic manure fertilization and lime. However, As-contaminated water use irrigated for crops, which decreased soil pH at the experimental end due to the H^+ number added to the farmland from As-contaminated water, released H^+ ion in the soil from soil acids into a free form (Van, 2024). Application of organic manures combined with chemical fertilizers raised significantly soil pH compared to the control treatment (Cuong & Chuong, 2022). Ensuring optimal plant growth and fertilizer utilization in As-contaminated soils, organic manures should be added to mitigate the harmful effects of heavy metals (Trang & Nguyen, 2024). Supplementation of organic manures is crucial to achieve the ideal soil pH for crop cultivation. Regular application of well-decomposed organic manures and NFM inoculation to As-contaminated soils effectively enhances soil buffering capacity and prevents abrupt pH fluctuations (He et al., 2022). Additionally, it improves phosphorus uptake and Fe and Al reduction in acidic soils (Chuong et al., 2024a). Application of animal manures and other organic fertilizers combined with NFM inoculation can be employed to boost plant yield, improve farmland fertility and acid soils. Precise determination of fertilizer application rates is essential to enhance soil pH, soil fertility, and plant productivity in As-contaminated soils. The integrated use of NFM inoculation with organic manures and inorganic fertilizers is officially considered a promising approach for sustainable crop production on As-contaminated soils (Chuong, 2024b). The results presented in Table 2 are consistent with the findings of the previous study by Ding et al. (2024); Nguyen (2023), who suggested that the vermicompost application combined with Bacillus vireti supplementation and RW irrigation (non-As-contaminated water) increased plant height taller than without vermicompost application, without Bacillus vireti supplementation, and UW (As-contaminated water) irrigation. The shoots developed into the ground to form pods, so if the plant has many available branches, it will produce more flowers and fruit (El-Sherbeny et al., 2023). The soil has a high sand content, resulting in poor water and nutrient retention. Combined with prolonged drought due to water scarcity and nutrient deficiencies, this poses a significant challenge. However, the application of endophytic bacteria strains in combination with earthworm manure has demonstrated significant improvements in these factors post-harvest (Chuong, 2024a). As shown in Table 3 could be attributed to the advantage relationship between vermicompost application, Bacillus vireti supplementation and RW irrigation with soil nutritional addition (Mathenge et al., 2019). The growth and biomass of peanuts increased significantly when the animal manure application combined with NFM inoculation (Jain et al., 2017). However, the complex effects of microbial diversity and rhizobia inoculation on crop yield of different peanut cultivars under organic and conventional farming systems is very limited (Zhang et al., 2024; Van Chuong, 2024). The results of the study were significant two-way interaction between farming types and factors that have beneficial effects on the quality and yield of peanut plants, such as earthworm fertilizer, nitrogen-fixing bacteria strains, and water for irrigation that is not contaminated with heavy metals (Paudel et al., 2023; AbdelRahman, 2023). Table 3 shows the significant interaction between three factors (vermicompost, *Bacillus vireti* supplementation and irrigation water types) at the 1% level. The previous studies discovered that organic manures fertilization coordinated with NFM inoculation raised the peanut yield components and yield (Trang & Chuong, 2025; Vadthe & Umesha, 2022). From these research results, all properties of peanut yield produced higher than where organic manures and were insignificantly applied. Application of vermicompost alone or combined with NFM inoculation were higher weight of 1,000 seeds of peanut than control (Chuong, 2025; Doloum et al., 2017). Their interaction of the experimental factors on peanut yield was insignificantly different at the 5% level (Table 4). The significantly negative correlations among the number of shoots, root biomass, and nodule number with some other traits, especially without organic manures application, non NFM inoculation, and heavy metals - contaminated irrigation water. These negative correlations could be due to decreased root and shoot biomass of peanuts in conventionally cultivated fields. However, application of inorganic fertilizer combined with organic fertilizer, NFM inoculation, and non-contaminated irrigation water, which help to increase the root and shoot biomass of peanuts, led to release readily available nutrients to the plants. Studies on other crops have shown that the use of chemical fertilizers combined with organic fertilizers has contributed significantly to increasing crop yield and quality (Chuong et al., 2024b; Ahmad et al., 2022). The soil As content decreased significantly, applying organic fertilizers to peanut plants, due to the lower As content in the organic manures compared to the soil, which reduced As uptake by peanut tems and seeds. Continuous application of clean organic manures can

be used to reduce the risk of As contamination in heavily contaminated soil-plant systems (Liu et al., 2024; Wang et al., 2021). The regulatory mechanisms of Bacillus vireti species involved in As absorption and accumulation in peanut plants that are still not clearly established. Therefore, recent studies have focused on the physiological and molecular mechanisms involved in the absorption, translocation, resistance, and detoxification of As by plants at different ABA concentrations or between different NFM trains. The results showed that Bacillus vireti has the ability to control As absorption and accumulation in plant tissues by appropriate inoculation of Bacillus vireti species in peanut cultivation (Jiang et al., 2023; Shahzad et al., 2023).

***Conclusion*:** The results of this study can be concluded that the application of 10 t vermicompost per ha coordinated with *Bacillus vireti* supplementation and RW irrigation (no As contamination) attained the highly positive effects on soil fertility, agronomic attributes, yield components, quality, and As accumulation in peanut stems and seeds. The application of 10 t vermicompost/ha coordinated with *Bacillus vireti* supplementation and RW irrigation increased peanut yields by 11.0, 14.3, and 23.4%, respectively, compared to the control. In particularly, the As concentration of the peanut stems and seeds reduced by 11.0, 12.9, 23.4%, and 12.9, 15.7, and 30.2%, respectively, compared to no vermicompost application, no *Bacillus vireti* supplementation and As-contaminated water irrigation (UW). From these results, it can be concluded that the combined application of vermicompost, *Bacillus vireti* supplementation, and non-As-contaminated irrigation increased both higher soil fertility, yield, quality and reduced lower As uptake in peanuts than those of control.

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